Hydrazones and their derivatives. II.* Preparation of pure D-erythrose

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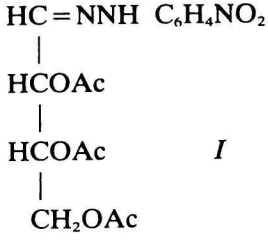
A method for preparation of pure D-erythrose is described in the paper. D-Erythrose can be separated from admixtures by treatment with (4-nitrophenyl)hydrazine under the formation of crystalline D-erythrose (4-nitrophenyl)hydrazone, from which pure D-erythrose can be liberated with benzaldehyde. 2,3,4-Tri-*O*-acetyl-aldehydo-D-erythrose (4-nitrophenyl)hydrazone can be obtained by acetylation of D-erythrose (4-nitrophenyl)hydrazone.

В работе описывается приготовление чистой D-эритрозы. D- Эритрозу можно от примесей изолировать реакцией с (4-нитрофенил)гидразином с образованием кристаллического (4-нитрофенил)гидразона D-эритрозы из которого можно выделить реакцией с бензальдегидом чистую D-эритрозу. Ацетилированием (4-нитрофенил)гидразона D-эритрозы можно приготовить 2,3,4-три-*O*-ацетил--альдегидо-D-эритроза (4-нитрофенил)гидразон.

Hydrazones of monosaccharides are generally well crystallizing compounds. They are also of interest because they exhibit several biological activities. D-Erythrose (4-nitrophenyl)hydrazone belonging to this group of compounds is known as an inhibitor of growth of some pathogenic yeasts [2]. Acetylation of D-erythrose (4-nitrophenyl)hydrazone gives 2,3,4-tri-*O*-acetyl-aldehydo-D-erythrose (4-nitrophenyl)hydrazone (*I*). Its acyclic structure was proved by mass spectrometry [1]. The compound *I* exhibits inhibitory effects on the synthesis of nucleic acids



* For Part I see Ref. [1].

and proteins in tumor cells [3, 4]. The present work is concerned with the preparation of the above-mentioned derivatives of D-erythrose and purification of D-erythrose *via* its (4-nitrophenyl)hydrazone.

D-Erythrose is a syrupy compound undergoing decomposition on a long-term storage. The decomposition is considerably accelerated already in the presence of traces of foreign substances. The presence of bases catalyzes aldolization [5], however, enolization and transformation of D-erythrose also can take place [6, 7]. The presence of acids initiates its dehydration [8].

One of the criteria for purity of D-erythrose is the value of its specific rotation. For instance, the Ruff degradation of D-arabinose [9] gives D-erythrose having specific rotation of $-19.1°$ At present, D-erythrose is considered for sufficiently pure when its specific rotation is $-38 \pm 3°$ [10]. The purest preparation of D-erythrose having $[\alpha]_D$ $-41°$ has been obtained by hydrolysis of 2,4-*O*-ethylidene-D-erythrose [11]. The most convenient route for the preparation of D-erythrose is the oxidation of D-glucose with lead tetraacetate [12]; the product prepared in this way is never sufficiently pure (specific rotation $-30°$).

The present work describes procedures for purification of D-erythrose from admixtures of other substances. They are based either on preparation of crystalline D-erythrose (4-nitrophenyl)hydrazone [13] or on a separation on a cellulose column [14]. Both procedures can be advantageously applied for D-erythrose

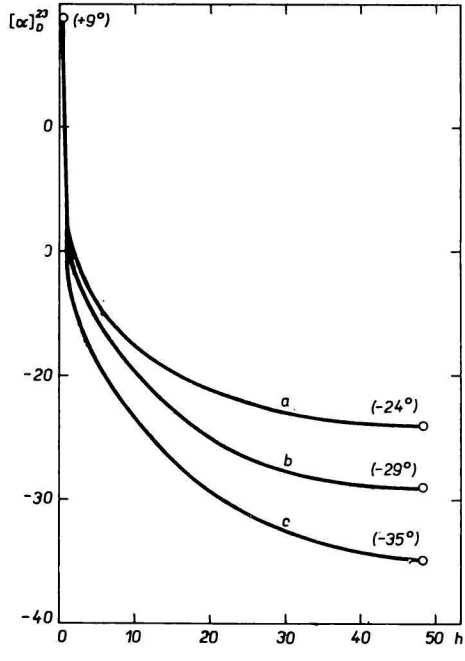


Fig. 1. Specific rotation of D-erythrose as a function of time and concentration. Concentration of D-erythrose in water: *a*) 9.1 %; *b*) 4.8 %; *c*) 1—2 %.

prepared by oxidation of D-glucose with lead tetraacetate. Crystalline D-erythrose (4-nitrophenyl)hydrazone is sufficiently stable and on treatment with benzaldehyde liberates pure D-erythrose; its specific rotation is a function of concentration and time (Fig. 1). Further drying over P_2O_5 of thus obtained D-erythrose gives anhydrous D-erythrose having specific rotation $-37.7°$

The preparation of chromatographically homogeneous D-erythrose is usually associated with difficulties, since even a pure product gives two spots after paper chromatography in several solvent systems. The ratio of the two fractions is dependent on the way of preparation of D-erythrose [12]. D-Erythrose gives only one spot after chromatography in the system ethyl acetate—acetic acid—4% boric acid in water which is also suitable for preparation of D-erythrose on a larger scale. Purification of D-erythrose prepared from D-glucose by oxidation with lead tetraacetate on a cellulose column eluted with the above solvent system gives chromatographically homogeneous product having optical rotation $-42°$

Experimental

Melting points were determined on a Kofler stage. Solutions were evaporated under reduced pressure at 30—40°C. Specific rotations were measured with an automatic polarimeter Bendix—Ericsson. D-Erythrose was prepared by oxidation of D-glucose with lead tetraacetate [12].

Descending paper chromatography was done in the system ethyl acetate—acetic acid—4% boric acid in water (9 : 1 : 1). Compounds were visualized with the periodate—benzidine reagent or with the aniline—hydrogen phthalate reagent. Preparative chromatography was carried out on a Whatman cellulose column (90 × 4.5 cm).

Preparation of D-erythrose (4-nitrophenyl)hydrazone

A mixture of D-erythrose (3 g) and (4-nitrophenyl)hydrazine (3.8 g) in ethanol (300 ml) was refluxed for 10 min and then left to stand at 4°C for 16 hrs. The formed crystalline product was filtered off, washed with ether (2 × 50 ml), and dried. Resulting D-erythrose (4-nitrophenyl)hydrazone (4.8 g; 75.3%), m.p. 209—211°C (Ref. [7] m.p. 212—214°C), could be directly used for preparation of pure D-erythrose.

In case D-erythrose (4-nitrophenyl)hydrazone was not sufficiently pure, it was recrystallized from a mixture ethanol—ethyl acetate. D-Erythrose (4-nitrophenyl)hydrazone (3.4 g; m.p. 202—205°C) was dissolved under heating in a mixture of ethanol (96%; 360 ml) and ethyl acetate (40 ml), the solution was concentrated to 150 ml and left to crystallize at 4°C to give D-erythrose (4-nitrophenyl)hydrazone (2.9 g; 85.3%), m.p. 209—211°C.

Preparation of D-erythrose from its (4-nitrophenyl)hydrazone

A mixture of D-erythrose (4-nitrophenyl)hydrazone (1 g), benzaldehyde (0.7 ml), benzoic acid (0.1 g), and water (40 ml) was refluxed for 45 min, then cooled to 10°C and at this temperature filtered to remove insoluble benzaldehyde (4-nitrophenyl)hydrazone which was washed with water (10 ml;

10°C). The filtrate was extracted with ether (5 × 20 ml), treated with charcoal, and evaporated. After drying the syrup to a constant weight in a vacuum desiccator over P_2O_5, pure D-erythrose was obtained (0.43 g; 92%), $[\alpha]_D^{25}$ −37.7° (*c* 1, water), (48 hrs).

Preparation of chromatographically homogeneous D-*erythrose*

D-Erythrose (5 g) was chromatographed on a cellulose column in the system ethyl acetate—acetic acid—4% boric acid in water (9:1 1). The effluent containing chromatographically homogeneous D-erythrose was evaporated and deprived of boric acid by successive evaporations with methanol. Resulting syrup was dried to a constant weight in a desiccator over P_2O_5 to give D-erythrose (4.5 g; 90%) having $[\alpha]_D^{25}$ −42° (*c* 1, water), (48 hrs).

*Preparation of 2,3,4-tri-O-acetyl-aldehydo-*D-*erythrose (4-nitrophenyl)hydrazone*

D-Erythrose (4-nitrophenyl)hydrazone (2 g) was added to a solution of acetic acid anhydride (4 ml) and pyridine (8 ml) cooled to 0°C. The mixture was first kept at 0°C for 2 hrs under occasional agitation, then at 3°C for 16 hrs. After removal of the acetylation mixture by distillation, ethanol (30 ml) was added to crystallize the corresponding acetyl derivative (2.6 g; 87%). Twofold recrystallization from ethanol gave product having m.p. 140—142°C and $[\alpha]_D^{25}$ +58.8° (*c* 0.5, ethyl acetate), (1 hr).

For $C_{16}H_{19}O_8N_3$ (381.34) calculated: 50.39% C, 5.02% H, 11.02% N; found: 50.48% C, 5.14% H, 10.92% N.

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